ASSESSMENT OF GENETIC VARIABILITY IN RESISTANCE TO BROWN SPOT NEEDLE DISEASE IN LONGLEAF PINE: ANALYSIS OF PERFORMANCE IN TEST CROSSES

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Brown-spot needle blight caused by the fungus *Mycosphaerelia dearnessii* (formerly *Scirrhia acicolia* (Dearn.)) is the most debilitating disease of longleaf pine (*Pinus palustris* Mill.). This pathogen attacks seedling foliage tissue causing cell death in sections of needles, or in severe infections, throughout entire needles. As a result, seedling vigor is reduced leading to delayed emergence from the grass stage, and in cases of heavy infection, seedling mortality. Given the serious negative impact on longleaf pine regeneration in areas where this disease is chronic, there has long been interest in mitigating its effects through resistance breeding (Kais 1975). Previous research has shown that genetic variability in susceptibility exists within extant longleaf pine populations and that breeding for resistance is likely to be successful (Derr and Melder 1970; Snyder and Derr 1975).

As a means to facilitate early screening of seedlings for susceptibility to this blight disease, artificial inoculation methods have been introduced for use in greenhouse environments (Kais 1975). If shown to produce resistance readings that correspond well with field observations, this methodology will be an indispensable aid to selecting families and possibly individual trees with improved resistance. In this report, we present preliminary findings from research designed to obtain additional information about genetic variation in resistance to this disease.

Materials and Methods

Progeny of 56 families produced by mating four male parents with 14 female parents in a factorial mating design were evaluated in greenhouse tests as well as at two high disease-hazard field sites, one on the Harrison Experimental Forest (HEF) in Harrison County, MS and the other on the Palustris Experimental Forest (PEF) in Rapides Parish, LA. Trees chosen for use as male parents included two that in earlier tests produced progeny with relatively high resistance to brown-spot disease as well as two that were observed to produce more susceptible progeny. The sample of female parents contained trees found to yield progeny spanning a range of resistance levels varying from moderate to high.

Artificial inoculations were administered under greenhouse conditions using techniques similar to those described in Kais (1975) and Lott et al. (2001). Two separate inoculation experiments comprised of seedling progeny were carried out in adjacent sections of a single greenhouse. Seedlings in one experiment were inoculated with spores cultured from longleaf needles collected from PEF, whereas seedlings in the second experiment were inoculated with spores from HEF needles. Each experiment consisted of 24 replications made up of seedlings from the entire complement of 56 full-sib families randomized in single tree plots. Following inoculation

and a full growing season in the greenhouse, surviving seedlings were transplanted to the field sites using the same experimental design employed in the greenhouse — seedlings inoculated with PEF spores were transplanted to PEF and seedlings inoculated with HEF spores were transplanted to HEF. Prior to transplanting, diseased foliage was removed from each seedling.

In this report we summarize results for brown-spot disease damage to seedling in the greenhouse experiments three months post-inoculation and brown-spot disease damage ratings and total height at the field sites after two and five growing seasons, respectively. Severity of brown-spot disease damage to trees was visually estimated as percent of total needle area killed by the fungus.

Results

Foliage damage scores were transformed using the logit function prior to statistical analysis; however means and standard deviations are presented here in terms of the untransformed scale. Generally, light to moderate disease damage was observed in the greenhouse tests (GH) (Figure 1); nevertheless mean damage to seedlings exposed to the PEF inoculum (15.5%) was 20 percent less than the mean value for seedlings treated with HEF inoculum (19.4%) ($p < 0.0001$). Such a difference suggests that the two inocula differ in pathogenicity toward the longleaf pine population that was the focus of this study. After two growing seasons, damage observed in both field tests (FY2) was higher than in the greenhouse tests (Figure 1). It is worthy of note however, that the difference was clearly much greater for the HEF test. Mean foliage damage was 154 percent higher at this field site than in the PEF test (48.8% vs. 19.2%). This large difference ($F = 15.969$, $p < 0.0001$) may have resulted from a number of factors, including differences between the sites in pathogenicity of their respective pathogen populations, in fungal spore loads, and possibly in weather conditions conducive to spore dispersal. Regardless of the cause, these experiments provided the opportunity to study family performances under both endemic and epiphytotic conditions.

Narrow-sense heritability values for damage scores estimated from greenhouse data were low for both inoculum sources ($h^2 < 0.1$, Figure 2). Nonetheless, evidence was detected for variation among male-by-female family effects ($p < 0.07$ for PEF inoculum, $p < 0.0001$ for HEF inoculum), which suggests, modest nonadditive genetic effects are involved in protecting seedlings from disease damage. A different but somewhat similar pattern was found for narrow-sense heritability for damage estimates made after two field seasons. At the PEF site, the estimated value ($h^2 = 0.25$) implies presence of moderate additive genetic variance for resistance to brown-spot disease damage, whereas the estimate for the HEF site ($h^2 = 0.09$) was much lower (Figure 2) suggesting that less additive genetic variability is present in this trait for conditions existing at this location. It is conceivable that the high levels of disease damage observed at HEF (Figure 1), resulted in reduced expression of genes that influence inhibition of disease spread. Even so, much like inferences drawn from the greenhouse tests, F tests for the presence of male-by-female interaction effects ($p < 0.003$ for PEF, $p < 0.07$ for HEF) indicate that nonadditive genetic effects are involved in limiting disease development.

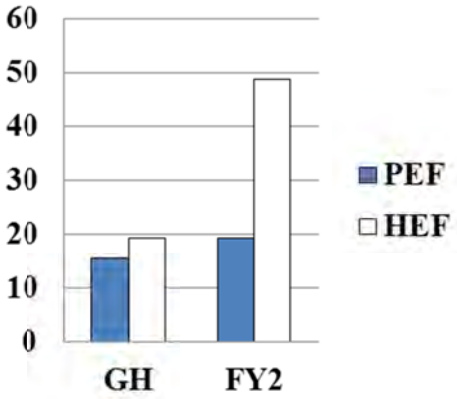

Figure 1. Means for percent foliage damaged.

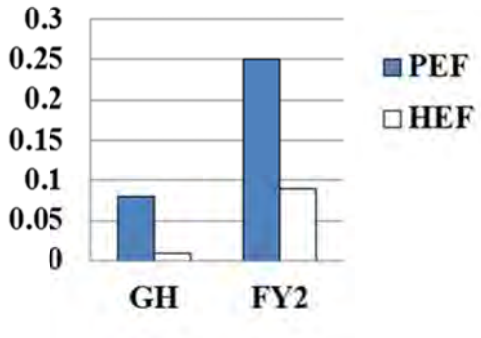

Figure 2. Narrow sense heritabilities for foliage damage scores.

Although there has long been awareness that increasing levels of brown-spot disease leads to decreasing tree height at intermediate stand ages (Boyer 1972), little is known about height growth decline in young stands. From a preliminary examination of our data, we noticed tree height variation also appears to decrease as foliage damage increases. Rates of decline for mean heights and standard deviations in trees at age five with increasing foliage damage at age two are illustrated in Figure 3. Similar patterns of growth loss and decreasing standard deviations were observed at the two test sites in our study. Mean height growth falls rapidly as foliage damage increases from ≤ 10 percent to 30-40 percent, then declines less steeply as foliage damage continues to climb. Standard deviation values drop less sharply, demonstrating that changes in this statistic are not a direct reflection of factors affecting the decline in mean height.

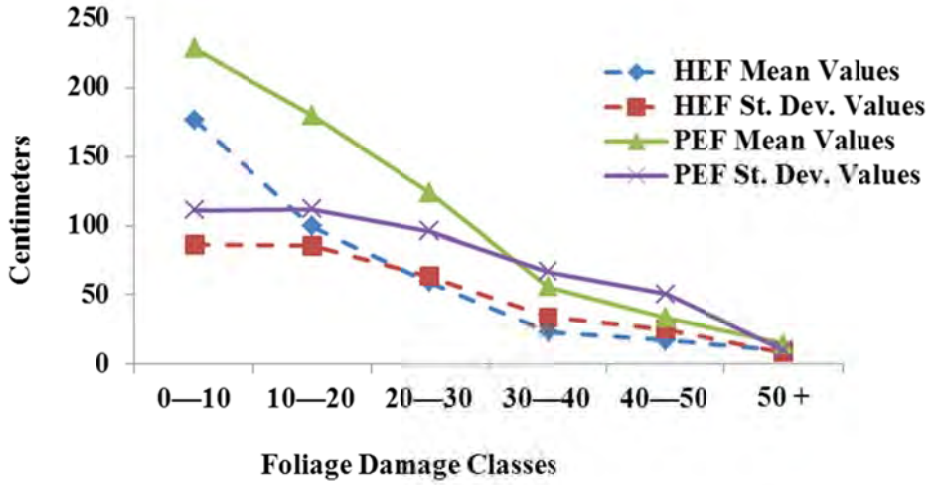

Figure 3. Changes in summary statistics for tree height at age five as foliage damage increases in two year old seedlings.

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